Optimization of scaffolds with *Buddleja globosa* Hope extract to prevent and treat chronic wound infections

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Introduction

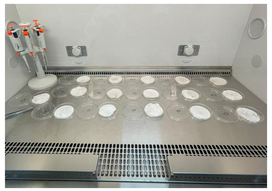
Wound infections negatively impact the healing process, and most chronic wounds are colonized by **bacterial biofilms** (1). *Buddleja globosa* Hope (BG) extracts have been traditionally used to treat skin wounds for their healing, anti-inflammatory, and antimicrobial properties (2). Scaffolds have been shown to promote cell proliferation and healing (3) and could be used to deliver bioactive compounds. However, the development of scaffolds rarely describes a rational design to optimize their therapeutic properties (4).

Objective

This work aimed to develop an optimized scaffold with BG extract by Design of Experiment (DoE) to prevent and treat wound infections.

Methods

Development of scaffolds

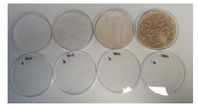


- First, 13 scaffold prototypes were prepared with variable %chitosan, %hyaluronic acid, and % gelatin (variables of the DoE) and fixed BG extract by lyophilization using a Box-Behnken design (BBD) (5).

Evaluation of scaffolds prototypes

- Bacterial adhesion and viability, and viability of mature biofilms were tested against biofilms *Pseudomonas aeruginosa* and *Staphylococcus aureus* in vitro.
- Compatibility was studied with human fibroblasts.

Optimized scaffolds



- Finally, 4 scaffolds with variable %BG extract and fixed polymeric content were developed and tested on bacterial adhesion, viability, and in a mature dual-specie biofilm model (6) in an artificial wound bed (7).

Results

Effect of the polymeric content

The BBD methodology showed that %chitosan correlated with the reduced viability of *S. aureus* (R^2= 0.98; p=0.0012) and reduced adhesion of *P. aeruginosa* (R2= 0.93; p=0.0195) (Figure 1).

Compatibility with fibroblasts correlated with %gelatin (R^2= 0.96; p=0.0064).

No correlation was observed between the model and the inhibitory activity against preformed biofilms of *P. aeruginosa* and *S. aureus*

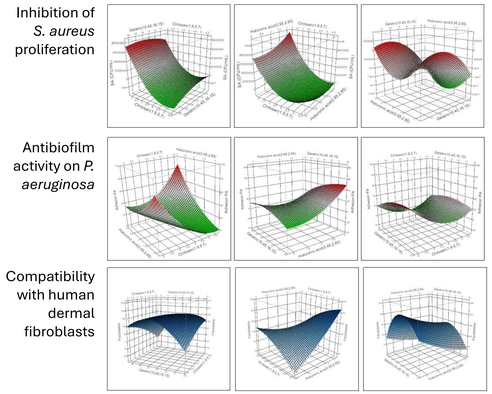


Figure 1. **Relation between polymeric content and both antimicrobial properties and compatibility** by a Box-Behnken design

Scaffolds on bacterial adhesion and viability

Optimized scaffolds with BG extract significantly reduced bacterial adhesion and viability (Figure 2) , and reduced the viability of mature dual-specie biofilms (p=0.0181).

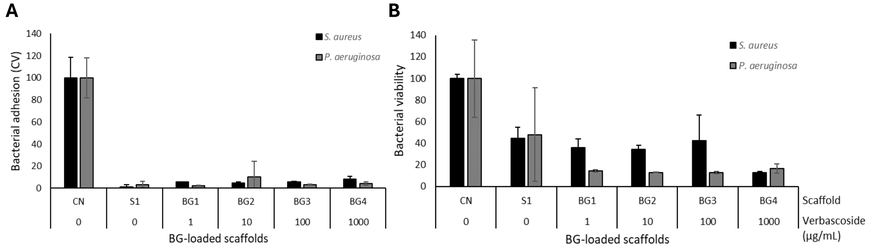


Figure 2. **Bacterial adhesion and viability after treatment with optimized scaffolds.** A) Bacterial adhesion to the well by cristal violet and B) Bacterial viability within scaffold by metabolic assays.

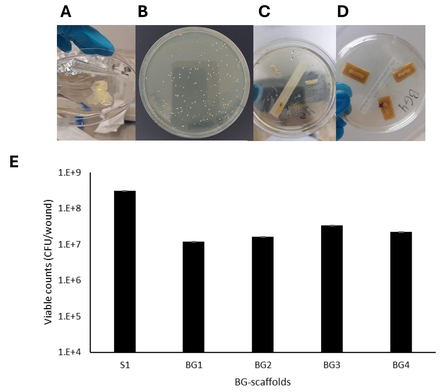


Figure 3. **Evaluation of scaffolds on a dual-specie biofilm *in vitro*.** Preformed biofilms of *P. aeruginosa* and *S. aureus* (0.5 g) (A) were placed in an artificial wound bed consisting of Bolton broth, 1% gelatin and 1.2% agar (C) for scaffold evaluation (D). B showed colonies of both species after dispersion of the biofilms and plating by spread plate method.

Conclusions

- **Scaffolds made of these polymers should balance their chitosan and gelatin content to potentiate the antimicrobial properties without compromising biocompatibility**
- **BG extract reduced the viability of bacteria entrapped in the scaffolds and the viability of bacteria in a dual specie biofilm model *in vitro*.**

Acknowledgements

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